(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) Publication number : **0 510 913 A2**

(12) **EUROPEAN PATENT APPLICATION**

(21) Application number : **92303554.7**

(51) Int. Cl.[5] : **A61K 37/30,** C07K 7/36

(22) Date of filing : **21.04.92**

(30) Priority : **23.04.91 GB 9108634**

(43) Date of publication of application :
28.10.92 Bulletin 92/44

(84) Designated Contracting States :
AT BE CH DE DK ES FR GB GR IT LI LU NL PT SE

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(54) **Pharmaceutical compositions comprising calcitonin.**

(57) The invention provides fibrillated calcitonin for use in treating calcium deficiency diseases.

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nin may be from 1 to 200 mg/ml, preferably from 5 to 100 mg/ml. When used as a dispersion of fragmented fibrils the concentration of calcitonin may be up to 50 mg/ml.

Preferred ranges for fragmented hCT fibril suspensions are from 3 to 10 mg/ml for nasal or oral solutions and from 0.5 to 3 mg/ml for injectable solutions.

The compositions of the invention have a prolonged hypocalcemic effect such that in calcitonin therapy the number of injections required, or doses to be taken can be reduced, compared to those needed when conventional calcitonin solutions are used.

The compositions of the invention may also contain viscosity-increasing swelling agents an/or sugars an/or other pharmaceutically acceptable additives. As viscosity-increasing swelling agents there may be used hydrophilic partially etherified cellulose derivatives, hydrophilic polyacrylates, polymethacrylates, polyethylene glycols, polyvinyl alcohols or mixtures thereof. Suitable compounds include methyl cellulose, hydroxypropylmethylcellulose, polyethylene glycol, dextran, which may have a molecular weight of 20,000 to 80,000, but preferably about 40,000, sugars such as sucrose, fructose, glucose, lactose, mannitol and trehalose, ethanol, serum albumin, lysozyme and preservatives such as benzalkonium chloride, benzethonium chloride and chlorhexidine diacetate.

The amount of additives used can vary and may depend on the intended use. For example for nasal or oral solutions, 0.5 to 10% by weight of additive may be used. In the case of injectable solutions, sugars, polyethylene glycols or dextran would be used as the additive, usually in amounts of 0.5 to 10% by weight.

The invention is illustrated in the following examples.

Example 1

200 µl of 0.001% acetic acid/water is added to 20mg hCT powder and solubilisation is performed using a vortex mixer for 1-2 min. The resulting solution is allowed to fibrillate for 1 hour. Over the hard turbid gel of hCT-fibril are added 4 ml of 0.001% acetic acid; then ultrasonication is performed for 2 min. Electron micrographs of negatively stained sonicated hCT fibrils show the presence of rods as in Figure 1a. The original hCT fibrils are shown in Figure 1b. The rods of sonicated hCT fibrils were, on average, 15 nm in diameter and 26-130 nm long.

Example 2

Following the procedure of Example 1, hCT fibrillated gels are formed from a solution of 20 mg hCT in 100 µl.0.1% acetic acid. 10ml water are then added and sonication performed as described in Example 1 with similar results.

Example 3

The product from Example 2 is diluted with 0.9% aqueous NaCl solution to give a solution having an hCT concentration of 5 µg/ml. This is injected intravenously into a hypocalcaemic rat model and compared with a similar injection of a solution of hCT made up in the conventional manner by dissolving hCT powder. The dosage in each case is 5 µg/kg body weight. The results are shown in Fig 2. The solution of Example 2 is biologically active and has a longer time dose response than the conventional solution.

Example 4

hCT fibrillated gels are formed by placing aqueous hCT solutions: (50 mg/ml hCT in 0. 1 % acetic acid or in 0.9% NaCl) in syringes and allowing the hCT to fibrillate in the syringes overnight. The sub-cutaneous (s.c.) intra-nasal (i.n.) intra-rectum (i.r.), intra-ileum (i.i.) or intra-colonical (i.c.) injection of hCT fibrillated gel can be done easily since the hardened fibrillated hCT gels can be squeezed through injection needles. Figures 3a and 3b shows that in the hypocalcaemic rat model s.c., i.n., i.r, i.i. and i.c. injected hCT fibrillated-gel have a strong and prolonged biological effect. Fibrils formed in 0.9% NaCl were used for s.c. and i.n. experiments and fibrils formed in 0.1% acetic acid were used for the i.r., i.i. and i.c. experiments.

In the in vivo experiments in Examples 3 and 4 the following hypocalcaemic rat model is used:-Female Wistar rats 80-100g body weight are fed a normal diet. hCT is administered as mentioned to animals anaesthetised with Hypnorm then sodium pentabarbitone i.m. Blood samples (250 µl) are taken from a cannulated carotid artery at each time point. Change in calcium level after the administration of hCT formulations is measured by a colorimetric method using a Ca measuring kit (Sigma Chem. Co.). For each time point three to five animals are used.

Claims

1. Fibrillated calcitonin for use in treating calcium deficiency diseases.

2. Fibrillated calcitonin as claimed in claim 1 which is human, salmon, eel or porcine calcitonin.

3. Fibrillated calcitonin as claimed in claim 1 or 2 which comprises calcitonin and water and optionally NaCl, a weak acid or a buffer.

4. Fibrillated calcitonin as claimed in any preceding claim which also comprises a viscosity-increasing gelling agent and/or a sugar and/or other pharmaceutically acceptable additive.

FIG.1a

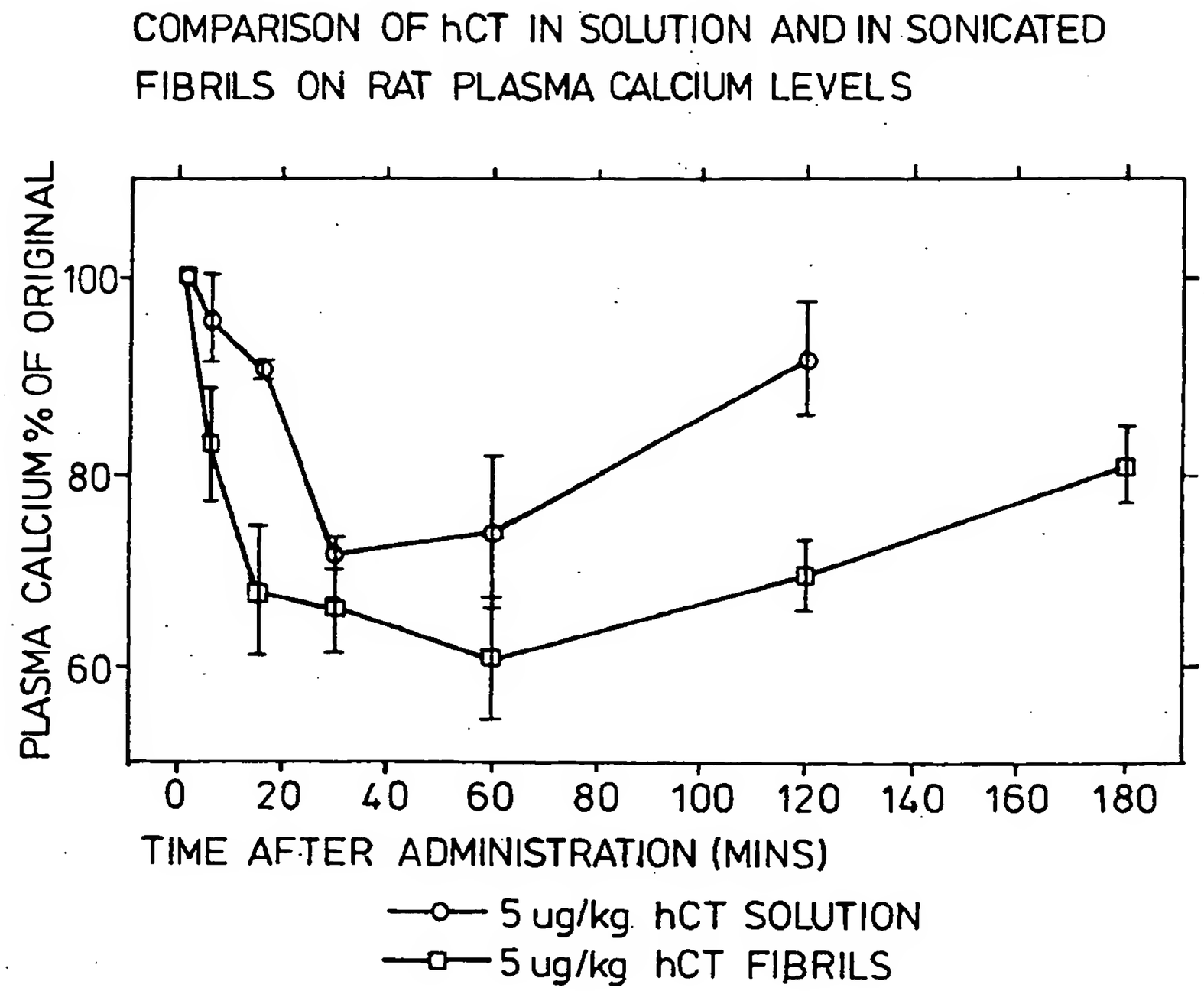
COMPARISON OF hCT IN SOLUTION AND IN SONICATED FIBRILS ON RAT PLASMA CALCIUM LEVELS
PLASMA CALCIUM % OF ORIGINAL
100
80
60
0
20
40
60
80
100
120
140
160
180
TIME AFTER ADMINISTRATION (MINS)
5 ug/kg hCT SOLUTION
5 ug/kg hCT FIBRILS

FIG. 2

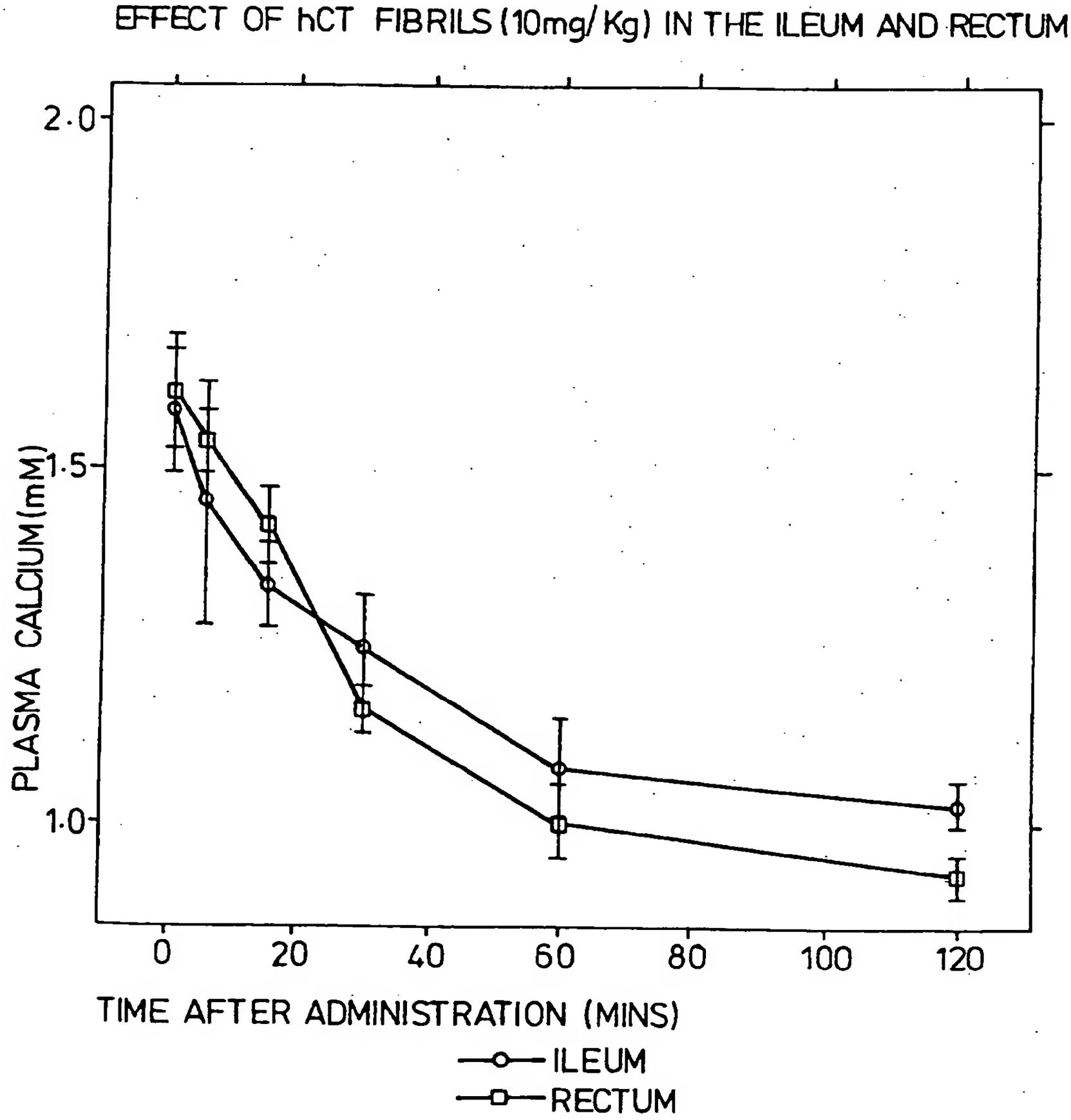
EFFECT OF hCT FIBRILS (10mg/Kg) IN THE ILEUM AND RECTUM
PLASMA CALCIUM (mM)
2·0
1·5
1·0
0
20
40
60
80
100
120
TIME AFTER ADMINISTRATION (MINS)
ILEUM
RECTUM

FIG. 3b